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THE KARYOTYPE OF THE DAMSELFLY, *LEPTAGRION MACRURUM* (BURMEISTER, 1839), AND ITS POSSIBLE ORIGIN, WITH A NOTE ON THE CYTOTAXONOMIC AFFINITIES OF THE GENUS (ZYGOPTERA: *COENAGRIONIDAE*)

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The diploid chromosome number of *Leptagrion macrurum* (Burm.) from the surroundings of Sao Paulo (Brasil), 2 n ♂ = 30, is the highest ever found in a dragonfly. Coupled with the neo-XY sex determination, it indicates the secondary nature of the karyotype. In Figure 2 a tentative model of the karyotypic evolution leading to the present peculiar situation is given. The cytotaxonomic evidence seems to be in favour of an ischnurine rather than pseudagrionine affinity of the *Leptagrion* alliance.

INTRODUCTION

On his study tour to South America, in October 1970, Prof.Dr. J.W. BOYES (Biology Department, McGill University, Montreal) made a series of lacto-acetic-orcein squash preparations of the testes of two specimens of *Leptagrion macrurum* (Burm.) collected at Boracea nr. Sao Paulo, Brasil. As often before, the author of this note has had the privilege to examine them cytologically, while Dr. D.C. GEIJSKES (State Museum of Natural History, Leiden) kindly identified the insects.

Though only six spermatogonial metaphase figures were present in our material, and spermatocyte stages, save for a few pachytene figures, were completely lacking, it soon became apparent that the cytological picture of the species is unique among the hitherto studied representatives of the Order.

It seems useful, therefore, to consider the karyotype in some more detail.

MORPHOLOGY OF THE KARYOTYPE

The diploid chromosome number, 2 n ♂=30, is the highest ever found in a dragonfly. The even number indicates a secondary mode of sex determination. The neo-sex elements are recognizable in all spermatogonial metaphase figures; the neo-X is by far the longest and the neo-Y the smallest element of the set at this stage. A part of the autosomal elements is nearly uniform in size and shape, the others are of gradually decreasing magnitude. The *m*-chromosomes are lacking (cf. Fig. 1).

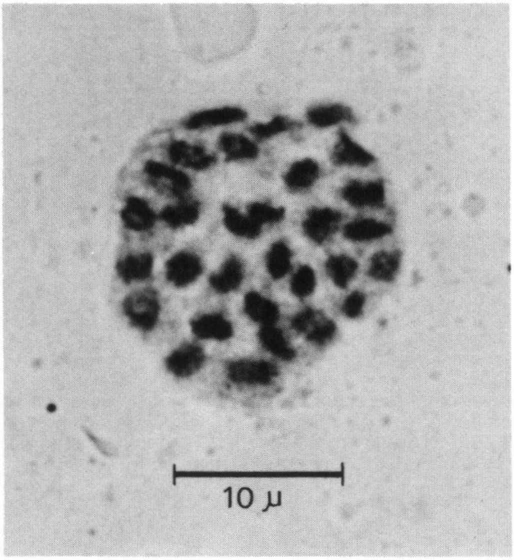

Fig. 1. *Leptagrion macrurum* (Burm.). (Sao Paulo, Brasil). Spermatogonial metaphase. (Lacto-acetic-orcein squash, 2250 X).

At pachytene no element is entirely heteropycnotic, though one, considered to represent the neo-X, is slightly less despiralised than the others.

THE ORIGIN OF THE COMPLEMENT AND THE EVOLUTION OF SEX DETERMINATION

Though nothing can be said with certainty as to the origin of the complement in *Leptagrion macrurum*, the situation in this species seems essentially similar to that found in the libellulidan *Neurothemis tullia tullia* (Drury) (RAY CHAUDHURI & DAS GUPTA, 1949), in which the original chromosome number

(2 n ♂=25) has also been increased by three elements and the original XO sex determination evolved into the neo-XY mode (cf. KIAUTA, 1969a). It can be tentatively assumed, therefore, that in *Leptagrion* too, the numerical increase in the complement is due to fragmentation of two autosomal pairs and subsequent fusion of the original X with a fragment of an autosome.

The fragmentation resulted in a diploid complement, 2 n ♂=31, in which the XO sex determination was still retained. The subsequent (or perhaps simultaneous) fusion of the original X with an autosomal fragment has probably reduced the chromosome number by one element in both *Neurothemis* and *Leptagrion* and has given rise to the neo-XY sex determination. The process is illustrated schematically in Figure 2.

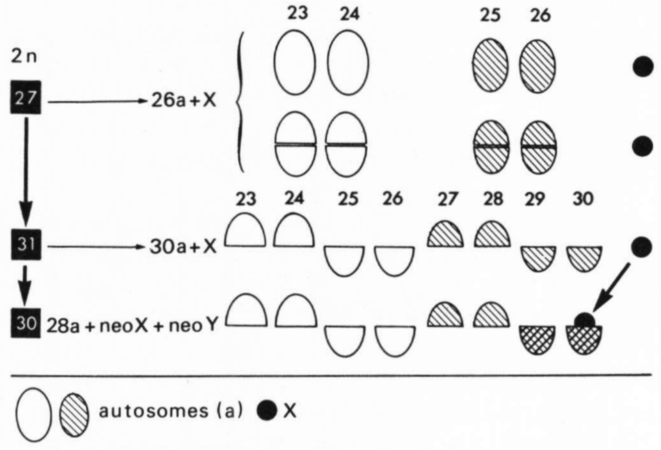

Fig. 2. *Leptagrion macrurum* (Burm.). Tentative interpretation of the increase of the chromosome number and the evolution of sex determination. Further explanation in text.

It should be stressed that this explanation remains speculative, since several other interpretations are also possible, while none can be proved sufficiently with our limited material.

DISCUSSION

The genus *Leptagrion* Selys, 1876, is endemic to South America; its type species is *Agrion macrurum* Burmeister. WILLIAMSON (1917) split off from the Selysian genus a new genus, *Aeolagrion*, with *Leptagrion dorsale* (Burmeister, 1839) as the type species.

Structurally, the two genera are well defined. Of particular interest are also their distinct ecological features. While most, if not all, *Leptagrion* species breed in water between the leaves of bromelias (cf. SANTOS, 1961, 1962, 1966, 1968a, 1968b; GEIJSKES, personal communication), the representatives of *Aeolagrion* seem to pass their larval life in small, seasonally nearly dried up pools and ditches of stagnant, or perhaps also very slowly running water (cf. WILLIAMSON, 1917).

The systematic position of the *Leptagrion* alliance within the family is far from being clear. FRASER (1957) placed *Leptagrion* in the *Pseudagrioninae*, but in his reclassification of the Order he did not make any reference to *Aeolagrion*. The latter was listed by CUMMING (1964) in the *Ischnurinae*.

It is unfortunate that only a single species of each genus has been studied cytologically. In *Aeolagrion foliaceum* (Sjöstedt) from Bolivia there are 14 elements in the male haploid set; none could be marked as an *m*-bivalent and sex determination is of the usual XO type (CUMMING, 1964).

So far the cytotaxonomic conditions in 52 members of the family have become known. They represent six subfamilies. The family type number is n = 14 (2 n ♂= 27) and the ischnurine *Enallagma cyathigerum* (Charp.) is the only hitherto known species where, in some populations, specimens or cells the chromosome number has been increased, by means of fragmentation, for one element (CRUDEN, 1968; KIAUTA, 1969b).

Though species with a pair of *m*-chromosomes are represented in most coenagrionidan subfamilies, a clear pattern seems to be detectable in the distribution of this feature. Thus, an *m*-bivalent is present in all examined members of *Amphicnemininae* (1) and *Pseudagrioninae* (11), it is lacking or uncertain in most but one *Coenagrioninae* (12) and is lacking in *Agriocneminae* (2) and *Argiinae* (4). In the subfamily *Ischnurinae* no *m*-bivalent was found in the members of the genera *Amphiagrion* (1), *Enallagma* (8), *Tigriagrion* (1) and *Zoniagrion* (1). It is present in *Acanthagrion* (1) and in one of the nine *Ischnura* species studied.

It is a matter of course, that the cytological observations alone do not suffice for systematic classification. The less so, since the sample of the taxa studied is too small to furnish reliable evidence. Nevertheless, it seems useful to indicate a few points:

(1) Aside from structural and ecological differences, the two representatives of the genera *Leptagrion* and *Aeolagrion* appear distinct also from the cytotaxonomic point of view. The recorded variation in the chromosome number on the species level, however, may be incidental rather than significant for the two genus groups.

(2) Of all coenagrionidan subfamilies, the increase in chromosome number has been found so far only in one ischnurine species. *Leptagrion* is the second case in the family. If, for other than cytological reasons, the *Leptagrion* alliance were to be classified under the *Ischnurinae*, the situation might well

indicate a peculiar cytogenetic feature of the subfamily. There are several cases in Anisoptera, where the tendency to an increase (or decrease) of the chromosome number in some well defined taxonomic units of different levels, represents a characteristic feature of their genetic system, e.g. the families *Gomphidae* and *Aeshnidae*, genera *Orthemis*, *Macrothemis* etc.

(3) The lack of an *m*-bivalent in the two genera certainly does not support the suggestion of a pseudagrionine character of the *Leptagrion* alliance. As far as this feature is concerned the two genera would certainly fit into *Ischnurinae*.

(4) It is, no doubt, only incidental that the second highest chromosome number in Zygoptera (2 n ♂=29) was found in a South American *Mecistogaster* (*Pseudostigmatidae*), i.e. in another form breeding in the bromelias.

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